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# Strategy for characterizing microbial physiology across scales in fermentation processes

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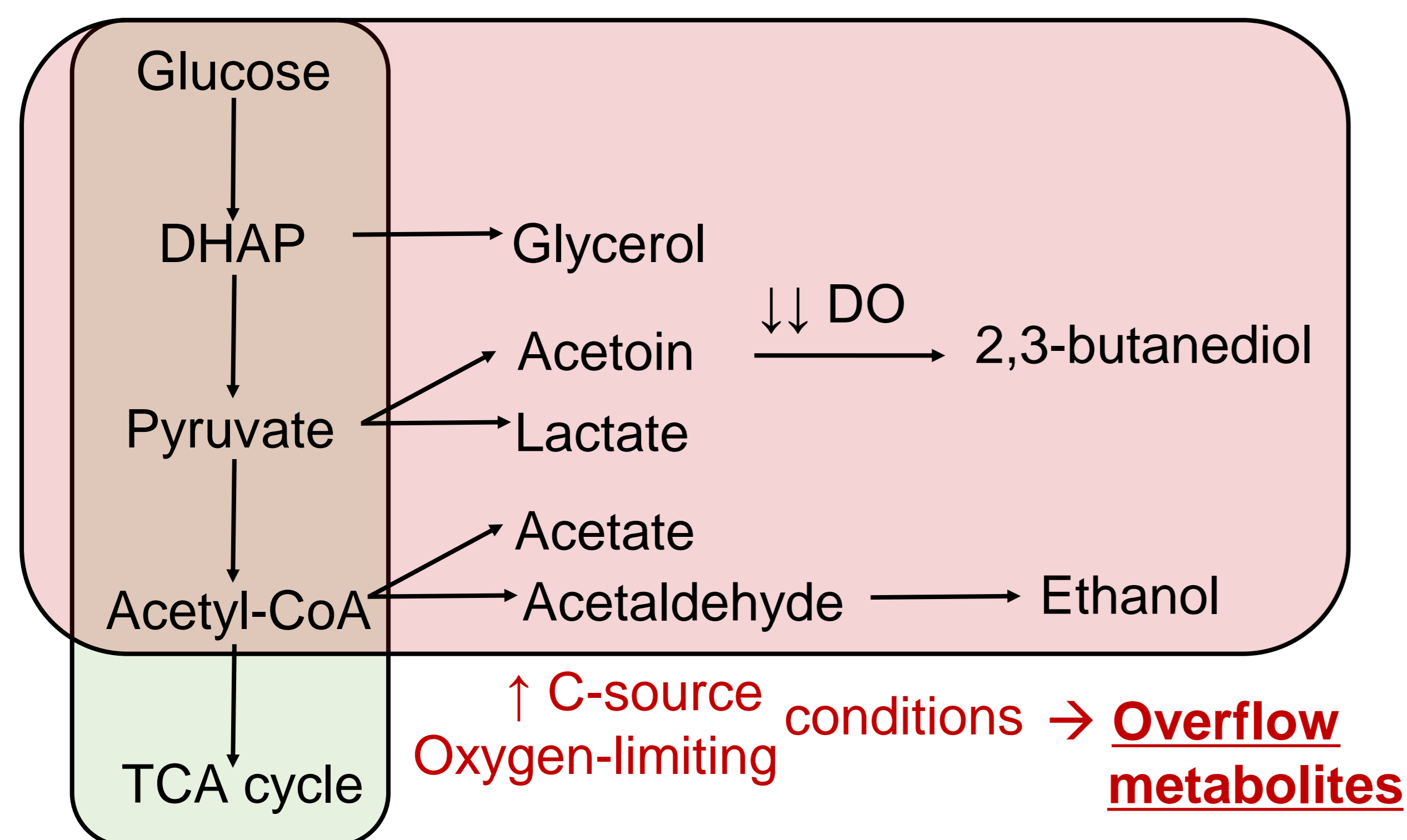
## Introduction

Industrial fermentation processes are **heterogeneous** because of limitations in the mixing and the mass transfer capabilities of the system. Thus, gradients in relevant reactor parameters such as **C-source and nutrient concentrations, pH, dissolved oxygen (DO) and dissolved carbon dioxide concentration** are likely to occur in large-scale [1]. As cells transition through the various zones of the reactor, they are constantly exposed to oscillatory conditions. Such oscillations can **affect cell physiology** at several levels. For instance, inducing stress responses, reducing yields and product quality [2] and causing changes in the cell metabolism and in the physiological properties of the cell [3]. Seeing the importance of physiology in the correct development of fermentation processes and its influence on productivity, this work presents a strategy to characterize gradients in different scales from a physiological point of view.

### Bacillus licheniformis as model organism

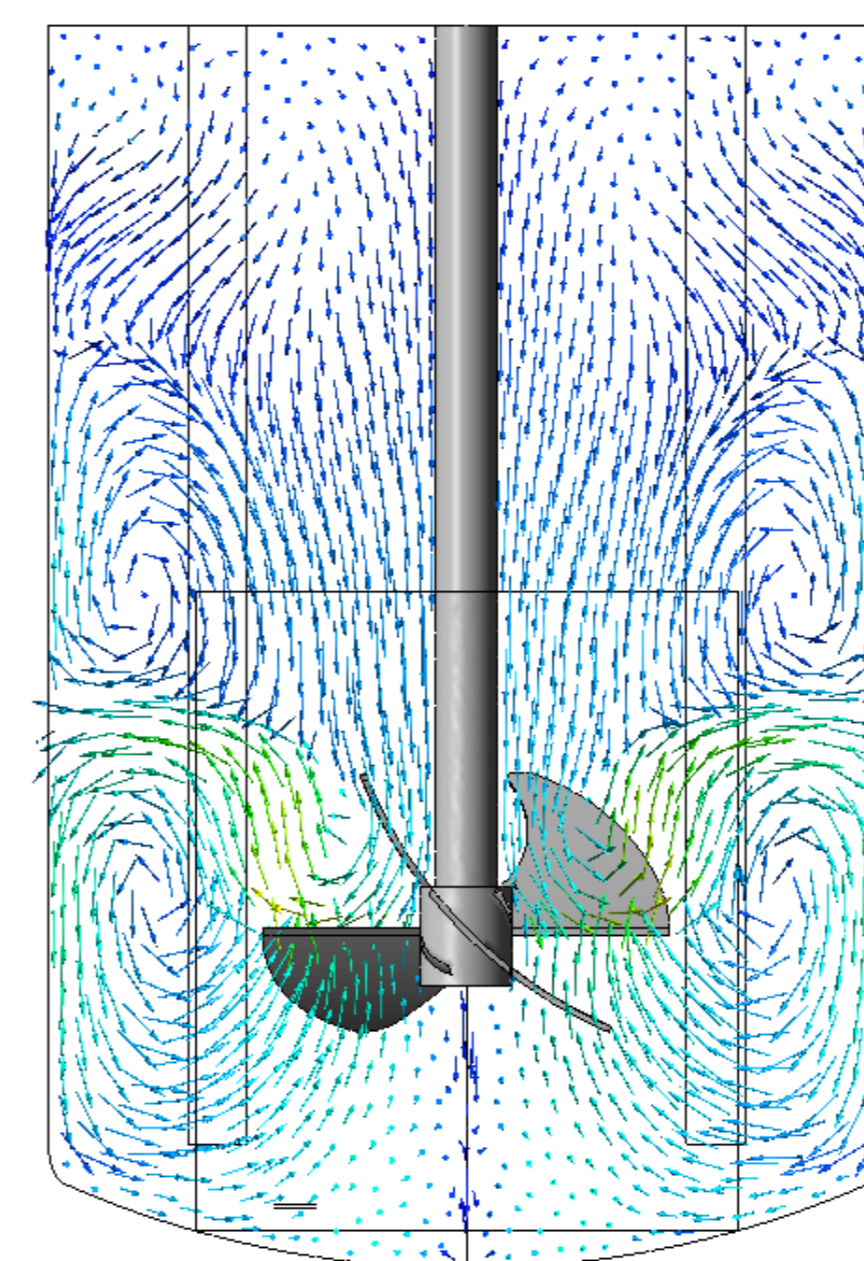
- ✓ Industrially relevant organism
- ✓ Sensitive to gradients – rapid metabolism
- ✓ Varied overflow metabolism in response to exceeding oxidative fluxes

#### Normal conditions



### Case study: glucose and oxygen gradients in an industrial fed-batch fermentation process

#### 1. Design of scale-down experiments using CFD



Geometry and mesh from [4]

#### Simulation of:

- ✓ Flow pattern
- ✓ Glucose and oxygen distributions
- ✓ Kinetics of *B. licheniformis*

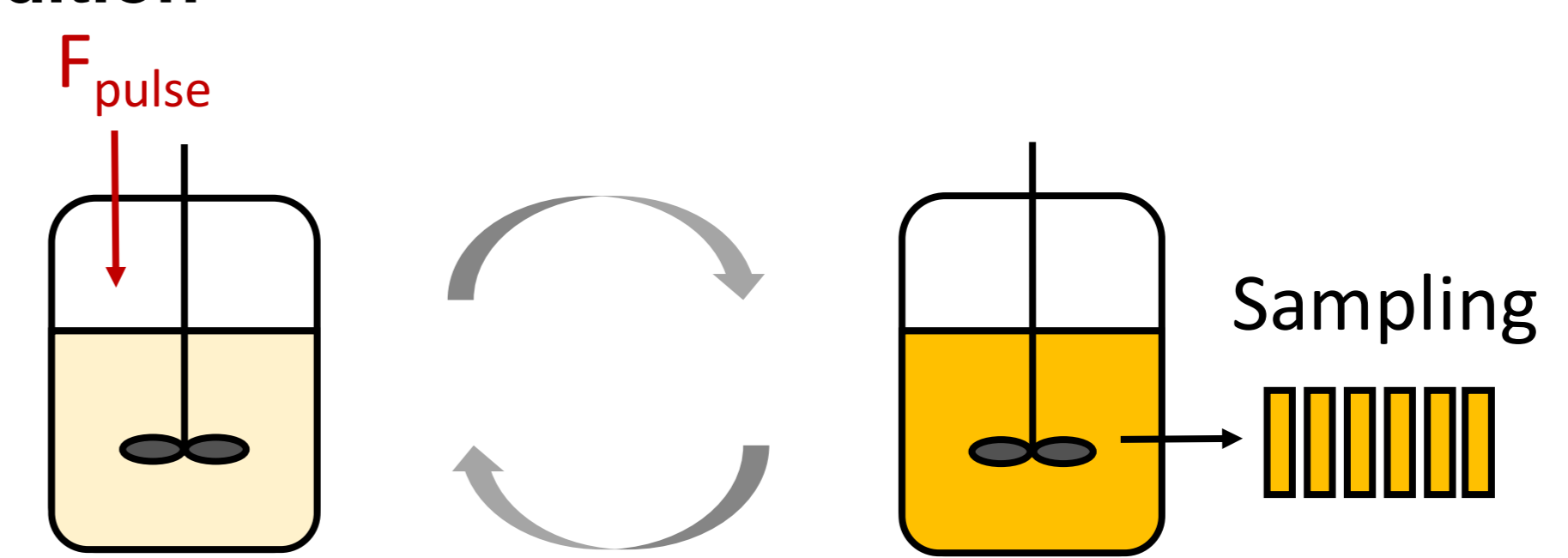
#### Outcome:

- ✓ Heterogeneous profile of substrate and product concentrations at several time points
- ✓ Starting point for scale-down experiments

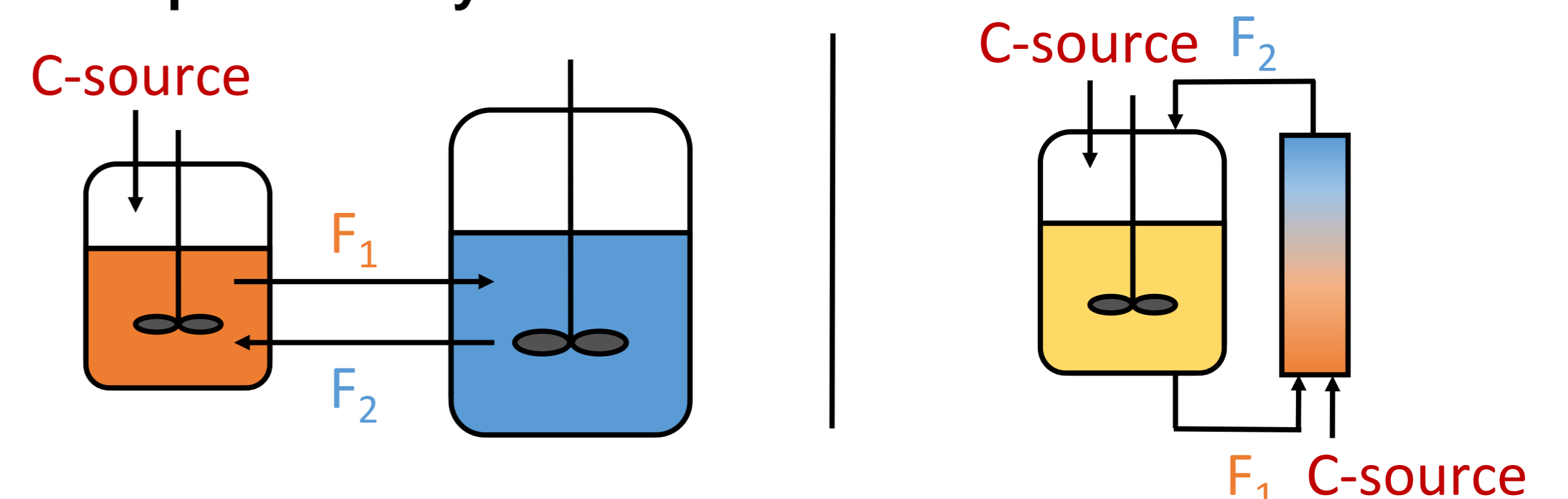
#### 2. Performance of scale-down experiments

Lab-scale systems in which microorganisms are subjected to temporal variations in given process parameters [5].

##### • Pulsed addition



##### • Multi-compartment systems

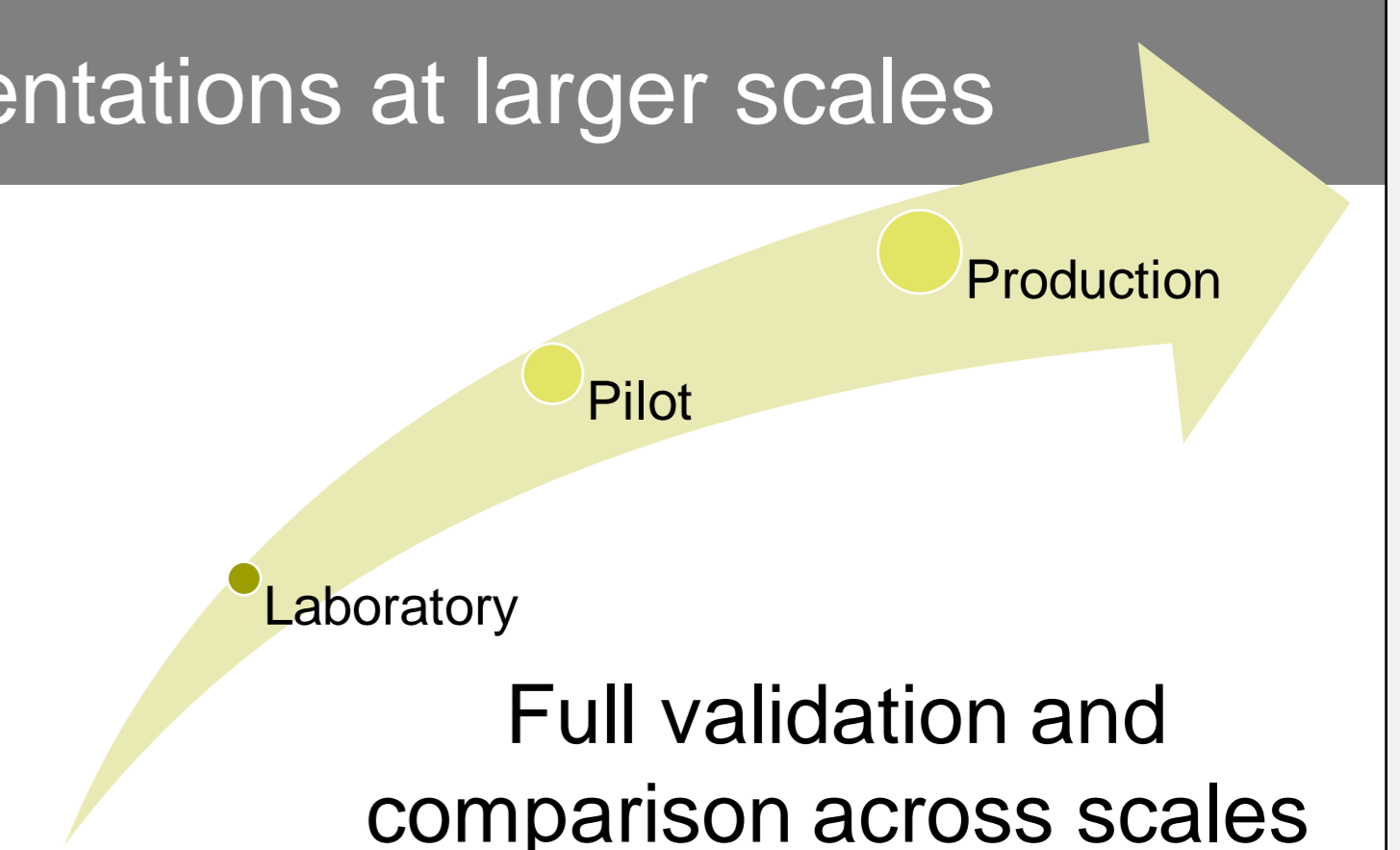


Seeing the results of the CFD model, one type or a combination of scale-down systems will be used.

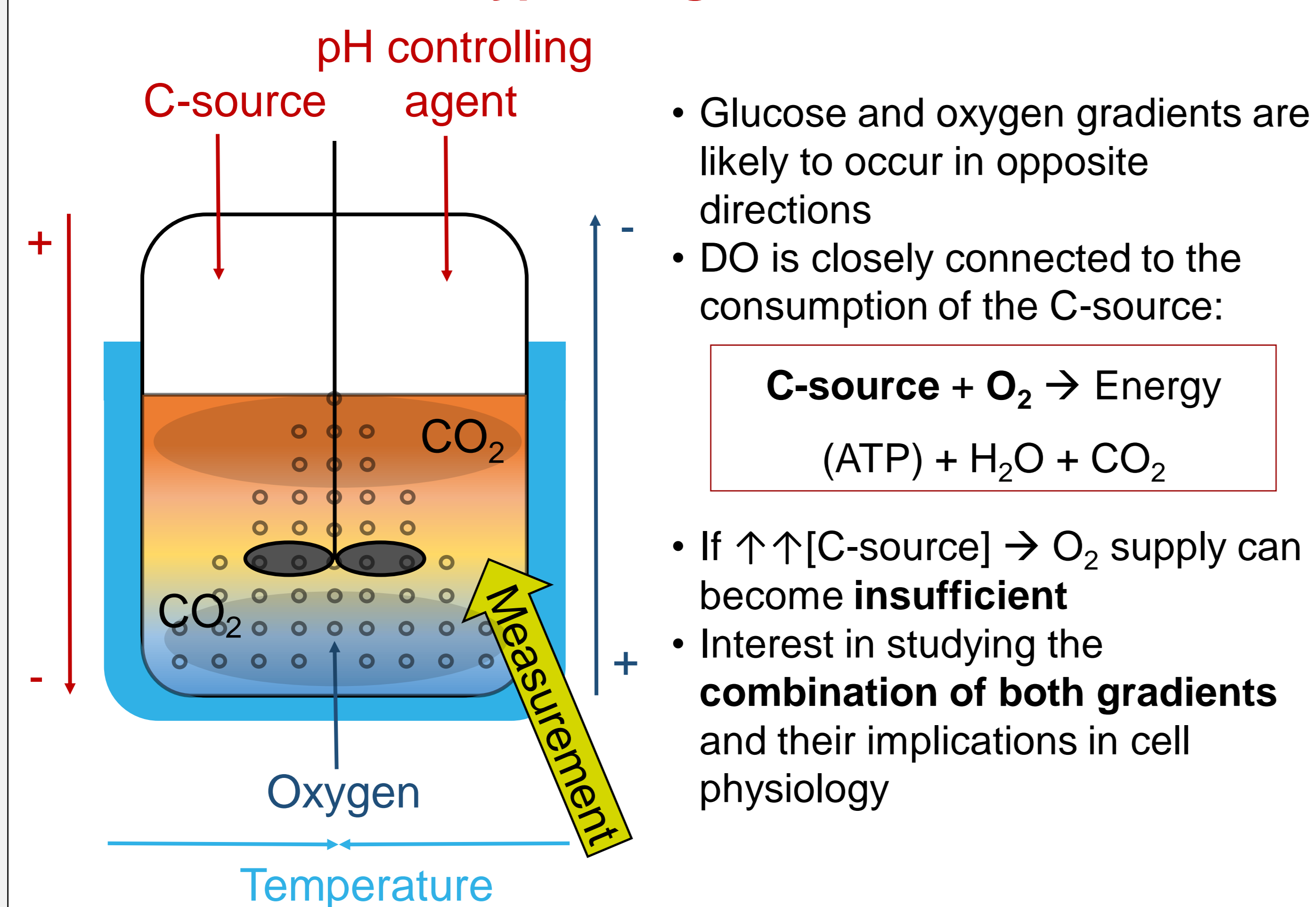
#### 3. Performance of fermentations at larger scales

Parameters to compare:

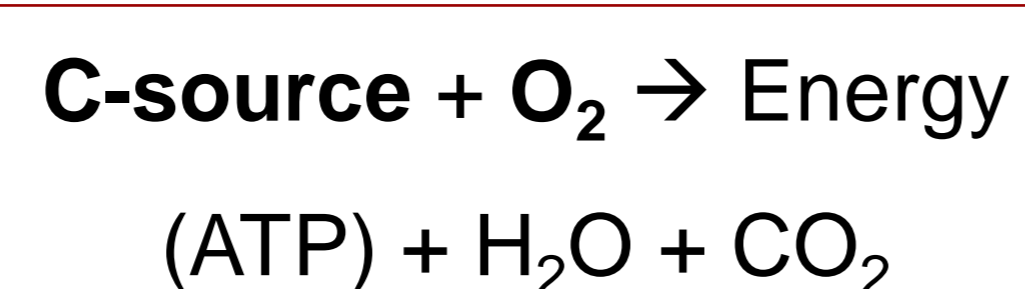
- ✓ Metabolic markers
- ✓ Productivity and product quality
- ✓ Cell physiological state



### Types of gradients



- Glucose and oxygen gradients are likely to occur in opposite directions
- DO is closely connected to the consumption of the C-source:



- If  $\uparrow\uparrow[\text{C-source}] \rightarrow \text{O}_2$  supply can become **insufficient**
- Interest in studying the **combination of both gradients** and their implications in cell physiology

### References

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